The opinion in support of the decision being entered today was not written for publication and is not binding precedent of the Board.

UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

MAILED

OCT 31 2006

U.S. PATENT AND TRADEMARK OFFICE
BOARD OF PATENT APPEALS
AND INTERFERENCES

Ex parte SVETLANA SHCHEGROVA, WILLIAM O. FISHER, and PETER G. WEBB

Appeal No. 2006-2664
Application No. 10/061,800

ON BRIEF

Before ADAMS, MILLS, and LINCK Administrative Patent Judges.

LINCK, Administrative Patent Judge.

DECISION ON APPEAL

This is a decision on appeal from the Examiner's final rejection under 35 U.S.C. § 103(a) of the pending claims 1-33 in Application No. 10/061,800 (hereafter the "'800 application").[1]

The invention on appeal relates to correcting errors in array fabrication by using redundant dispensers in place of error dispensers. There are three independent claims in

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[1] The present application was filed on January 30, 2002 and is assigned to Agilent Technologies, Inc.

the '800 application, claims 1, 6 and 25, from which all other claims depend. Claim 1 is the broadest claim and reads:

> 1. A method of fabricating a chemical array using:
> a head system with multiple groups of drop dispensers;
> a transport system to move the head system with respect to a substrate;
> a processor to dispense droplets from dispensers during operation of the transport system, in a pattern along a selected path for each group;
> the method comprising:
> a) loading the dispensers with fluid such that each dispenser group has at least one set of redundant dispensers loaded with a same fluid;
> b) dispensing drops from the dispensers to identify an error in one or more dispensers;
> c) moving a first dispenser of each set in each group along the selected path for that group while dispensing drops from non-error first dispensers of the sets in at least part of the pattern along the selected path for each group;
> d) moving a second dispenser of the sets in each group along the selected path for that group while dispensing drops from a non-error second dispenser of a set having an identified error first dispenser, in at least part of the pattern for the selected path of the first group; and
> e) repeating (a) through (d) at least once;
> wherein the array is fabricated.

Claims 1-3, 5-19, 21-29 and 31-33 of the '800 Application are rejected under 35 U.S.C. 103(a) as being unpatentable over Brown et al., U.S. Patent No. 5,807,522 issued Sept. 15, 1998 ("Brown"); and Tisone et al., U.S. Patent No. 6,063, 339 issued May 16, 2000 ("Tisone"). Additionally, dependent claims 4, 20 and 30 are rejected under 35 U.S.C. 103(a) as being unpatentable over Brown, Tisone, and Gamble et al., U.S. Patent No. 5,958,342 issued Sept. 28, 1999 ("Gamble").

With the authority to adjudicate appeals from final rejections under 35 U.S.C. § 134, we find that the Examiner has not established a prima facie case of obviousness. We reverse the Examiner's rejections of record.

BACKGROUND

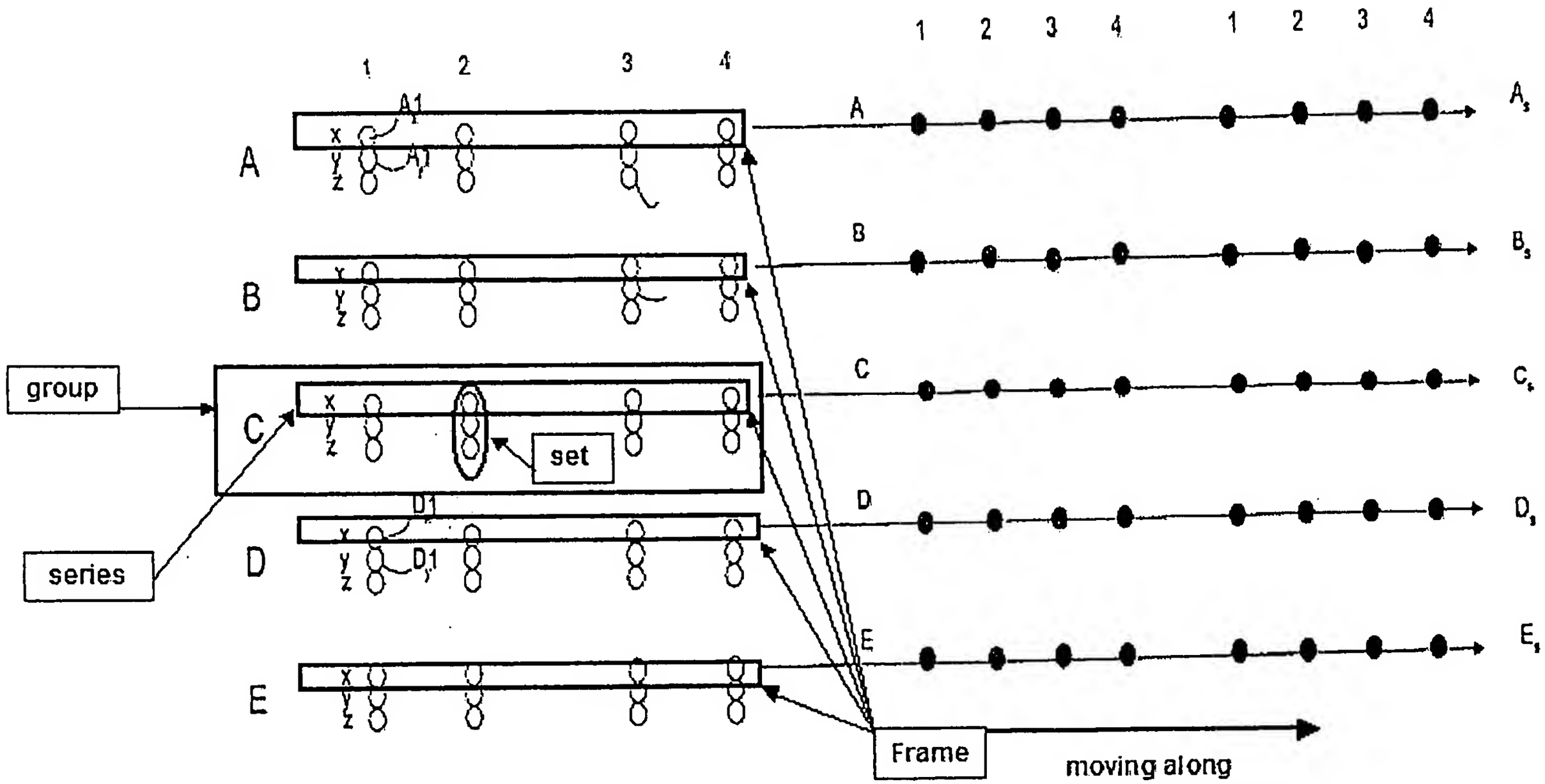
The invention generally "relates to arrays, particularly polynucleotide arrays such as DNA arrays, which are useful in diagnostic, screening, gene expression analysis, and other applications." Specification at 1. The inventors recognized a problem in array fabrications using multiple drop dispensers that move in relation to a substrate to deposit drops, where one or more dispensers may be in error. *Id.* at 3. The inventors also realized that "array quality can still be maintained by providing one or more redundant dispensers and an efficient way of using redundant dispensers in place of error dispensers." *Id.* "Dispensers of each set communicate with a common reservoir for that set" and, in effect, the dispensers of the same set are "loaded with the same fluid" and are "redundant." *Id.* at 12. During array fabrication, a functioning redundant dispenser ("non-error dispenser") is used in place of a previously identified error dispenser of the same set. *Id.*

Accordingly, the claims of the invention set forth methods that utilize a set of non-error redundant dispensers to correct identified error dispensers. The apparatus generally is comprised of a head system with multiple groups of dispensers, a transport system, and a processor. Specification at 3. Reproduced below, Figure 4B of the application shows an exemplary arrangement of "sets," "series," "groups," and "frames" of dispensers and a completed array (hollow circles represent drop dispensers and solid black circles represent deposited drops). The claim terms "sets," "series," "groups," and "frames" help identify particular groupings of dispensers in order to describe movement of dispensers according to the claimed method. Below in the figure, "groups" are marked

as A, B, C, D, and E and rows x, y, and z are "series" within each group. "[A] set of dispensers are those with the same group and column identification" (i.e. column 1, 2, 3, or 4). Specification at 12. For example, dispensers A_x1, A_y1, and A_z1 constitute a set. *Id.* A "frame" of dispensers is a

> shorthand way of designating series of dispensers from different groups of dispensers in a head system, which can simultaneously move along the selected paths for their groups forming a dispenser frame. For example, where the series are lines, the lines from each group which simultaneously move along the selected paths for their groups, form a frame. [*Id.* at 9.]

The methods claimed generally comprise the steps of loading each set of redundant dispensers with the same fluid; dispensing drops from the dispensers to identify an error; moving first dispensers or a frame of first dispensers along a selected path while dispensing only from non-error dispensers, and moving a redundant dispenser or frame with redundant dispensers along the selected path while dispensing drops from non-error redundant dispensers in the same set as the error first dispensers.



*** Note: the captions "group," "series," "set," "frame," and the corresponding arrows have been added for descriptive purposes. ***

FIG. 4B

DISCUSSION

Dependent claims 4, 20 and 30 each add a pulse jet limitation to the dispensers claimed in the aforementioned independent claims. We initially focus on claim 1, the broadest claim in the application.

Concerning the application of § 103(a), the Supreme Court has articulated three factors that are relevant to an obviousness determination: (1) the scope and content of the prior art; (2) the differences between the prior art and claims at issue; and (3) the level of ordinary skill in the pertinent art. *Graham v. John Deere Co.*, 383 U.S. 1, 17, 148 USPQ 459, 469 (1966). Thus, *Graham* instructs us to consider these three factors prior to turning to the ultimate legal conclusion.

The Scope and Content of the Prior Art

The invention's field of endeavor generally "relates to arrays, particularly polynucleotide arrays such as DNA arrays, which are useful in diagnostic, screening, gene expression analysis, and other applications." Specification at 1. The Examiner relies on two prior art patents (Brown and Tisone) as a basis for rejecting pending claim 1 under 34 U.S.C. § 103(a). Both references relate to the inventors' field of endeavor.

The Brown patent relates to a method and apparatus for fabricating microarrays of biological samples for large scale screening assays. Col. 1, lines 15-17. Brown discloses a "method of forming a microarray of [discrete] analyte-assay regions on a solid support, where each discrete region in the microarray has a selected, analyte-specific reagent . . . ." Col. 3, lines 24-27; col. 19, lines 1-4. Brown discloses a dispensing device (col. 7, lines 2-5), a transport system (col. 7, lines 31-33), a processor to dispense droplets

in a selected pattern (col. 10, line 63- col. 11, line 3), the step of loading a dispenser (col. 7, lines 55-58), and the step of depositing solution onto a surface by tapping the dispensing device against the support (col. 7, line 66 – col. 8, line 3).

Although the Brown reference discloses use of "multiple dispensing heads," (*see* Answer at 10), it does not disclose redundant dispensers as described and claimed by Appellants. Brown discloses that the "dispensing device in the apparatus may be one of a plurality of such devices which are carried on the arm for dispensing *different* analyte assay reagents at selected spaced array positions." Col. 4, lines 12-15 (emphasis added). Thus, the multiple dispensers in Brown are not loaded with the same fluid. Moreover, the Examiner concedes that Brown fails to teach the step of identifying an error dispenser. Answer at 5.

Tisone is the second reference relied upon by the Examiner to reject claim 1. The Tisone patent relates to "a method and apparatus adapted for high-speed, precision dispensing of high-density 'dot' arrays and other patterns onto a receptive membrane, high-density micro-well plate or other suitable receptacle." Col. 1, lines 13-16. Tisone discloses simultaneous dispensing from multiple dispensers of the same fluid in particular arrangements, i.e., in parallel or in another coordinated manner. Col. 7, lines 61-67; col. 22, lines 16-31. Tisone discloses an apparatus with a dispensing head (head system), a pump device, and a controller (processor). Col. 25, lines 40-67; col. 4, lines 22-28; col. 8, lines 18-25. Tisone also discloses that "*multiple dispensing heads* in linear or two-dimensional arrays can also be used with equal or improved efficacy" and "may be provided and operated either *in parallel* as illustrated in Fig. 2 (ie. for multi-gang

operation) *or in another coordinated fashion*, as desired," i.e. independent of one another. Col. 7, lines 61-67 (emphasis added); col. 22, lines 16-18. Furthermore, Tisone discloses the following:

> *Arrays of dispenser heads* could also be configured together... so as to provide array dispensing of 8, 16, or 64 drops *simultaneously* . . . Fig. 2 illustrates a single continuous feed platform . . . configured with *multiple dispensers* . . . *to handle one* or more *reagents*. This particular dispensing apparatus configuration has significant advantages for continuous web production applications since one or more syringe pumps . . . can be operated in alternating succession while allowing the non-dispensing syringe pump to draw additional reagent from the reservoir or they can be configured independent of one another *to dispense the same or different reagents simultaneously or in succession.*

Col. 22, lines 18-31 (emphasis added). These disclosures in Tisone teach or suggest groups, sets, series, or frames of dispensers and coordinated and simultaneous dispensing.

However, Tisone's disclosure of simultaneous dispensing of multiple dispensers of the same fluid in coordinated arrangements is *unrelated* to any error identification and correction. Tisone involves a method and apparatus for high-speed dot array dispensing that takes place "on-the-fly," i.e., "without the need to alternately stop and start the X-Y carrier platform." Col. 7, lines 35-39; col. 8, lines 25-27.[2] Thus, dispensing occurs while there is continuous motion between the substrate and dispensing head. Col. 4, lines 14-28.

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[2] The '800 Application invention is also "on-the-fly" dispensing because claim elements 1(c-d), 6(c-d), and 25 (c-d) recite steps of moving dispensers or frames *while* dispensing from dispensers. *See* claim 1.

"Phase adjustment," a type of error correction, occurs to "accommodate . . . on-the-fly dispensing without compromising accuracy, precision or repeatability." Tisone, col. 8, lines 25-28. The phase adjustment calculated for each dispense cycle "is such as *to advance (or retard) the timing of the valve opening and closing* so that the dispensed droplet of reagent . . . lands at the desired location on the substrate . . . (or at a desired offset location), taking into account its anticipated trajectory." Tisone, col. 8, lines 30-34 (emphasis added). Phase adjustments can be "determined experimentally . . . either before or during production" and

> will depend, among other things, on a number of system input and output parameters and behavioral characteristics, including the desired drop offset (if any), the vertical distance between the dispensing head nozzle . . . and the surface of the substrate . . . , the velocity and/or acceleration of the dispensing head . . . and/or the substrate . . . relative to one another, the velocity of the dispensed droplets, ambient temperature and humidity, and other controlled and/or uncontrolled factors.

Tisone, col. 8, lines 36-52.

Based on these disclosures, Tisone identifies errors with respect to where dispensed droplets land during on-the-fly dispensing and then corrects these errors by *adjusting* the parameters of the *same* error-dispensers. See, e.g., Tisone, col. 19, lines 6-9. Thus, Tisone fails to teach a method that utilizes the multiple, redundant dispensers for error identification and correction as recited in claim 1 elements 1(c-d), 6(c-d) and 25(c-d).

The Level of Skill in the Art

The level of skill in the art is not challenged and is reflected in the references cited in the case.

The Differences Between the Prior Art and the Claims At Issue

Appellants argue that the Examiner's prima facie case of obviousness is deficient because the combined teachings of the cited prior art fail to teach or suggest all the claim limitations of the rejected claims. Brief at 8. In particular, Appellants argue that Brown and Tisone do not teach or suggest at least the following features:

- "A head system with multiple groups of drop dispensers;"
- Claim element 1(a): the step of "loading the dispensers with fluid such that each dispenser group has at least one set of redundant dispensers loaded with a same fluid;"
- Claim element 1(b): the step of "dispensing drops from the dispensers to identify an error in one or more dispensers;"
- Claim element 1(c): the step of "moving a first dispenser of each set in each group along the selected path for that group while dispensing drops from non-error first dispensers of the sets in at least part of the pattern along the selected path for each group;" and
- Claim element 1(d): the step of "moving a second dispenser of the sets in each group along the selected path for that group while dispensing drops from a non-error second dispenser of a set having an identified error first dispenser, in at least part of the pattern for the selected path of the first group."

Id. at 9, 14, 15. Appellants describe their invention as a "method of fabricating an array [that] utilizes redundant dispensers (i.e., *Sets* of dispensers) in such a way that a drop that was not deposited by a first defective (or error) dispenser of a *Set* is deposited by a second (or third) non defective (or non-error) dispenser of the same *Set*." *Id.* at 11. The

Appellants state that this "configuration of dispensers makes the claimed method possible." *Id.* Essentially, according to Appellants, "Brown et al. and Tisone et al. fail to teach or suggest *Groups, Sets, Series,* or *Frames* of dispensers as is claimed" and "[w]ithout such a teaching, these references simply cannot teach the error correction array fabrication methods of the claimed invention." *Id.* at 19.

Furthermore, Appellants argue that Tisone fails to teach the steps of identifying an error dispenser, withholding dispensing from the error dispenser, and "dispens[ing] fluid from a *second* (or third) non-error (i.e., functional) dispenser selected from the same redundant *Set* in which the error dispenser is found." *Id.* at 14.

We agree with the Appellants' assessment of the differences between the prior art and the claims at issue in that Brown and Tisone, combined, fail to teach a method of error identification and correction that utilizes redundant dispensers as required by claim 1 steps (a)-(d).

The § 103(a) Determination in View of These Graham Findings

The issue before us is whether the evidence of record supports the Examiner's prima facie case of obviousness. In order to establish a prima facie case of obviousness, there must be "some objective teaching in the prior art or . . . knowledge generally available to one of ordinary skill in the art [that] would lead that individual to combine the relevant teachings of the references." *In re Fine,* 837 F.2d 1071, 1074, 5 USPQ2d 1596, 1598 (Fed. Cir. 1988). In some circumstances, a single prior art reference can render a claim obvious if there is "a showing of a suggestion or motivation to modify the teachings of that reference to the claimed invention in order to support the obviousness

conclusion." *SIBIA Neurosciences, Inc. v. Cadus Pharm. Corp.*, 225 F.3d 1349, 1356, 55 USPQ2d 1927, 1931 (Fed. Cir. 2000). Evidence of a suggestion, teaching, or motivation to combine or modify may flow from the prior art references themselves, the knowledge of one of ordinary skill in the art, or from the nature of the problem solved. *In re Kahn*, 441 F.3d 977, 987-88, 78 USPQ2d 1329, 1338 (Fed. Cir. 2006); *SIBIA Neurosciences*, 225 F.3d at 1356, 55 USPQ2d at 1931.

The invention's error correction method requires step b, the identification of error dispensers; step c, dispensing only from non-error dispensers on the initial pass; and step d, on subsequent passes, dispensing from *redundant* non-error dispensers loaded with the same fluid as the error dispensers that did not function on the first pass. These three steps require the loading of redundant dispensers (step a). None of these steps is disclosed by the cited prior art.

With respect to claim 1, the Examiner applies Brown as being directed to an apparatus with a positioning structure (transport system), a dispensing structure (head system) with a dispensing device for depositing a fluid onto the surface of the substrate, and a control unit (processor) that controls the positioning and dispensing; and a method comprised of loading the dispenser with a reagent solution, moving the dispenser to a selected position with respect to a support surface, dispensing the solution reagent onto the surface of the substrate, and repeating the steps to produce an array. Answer at 4-5 (citing Brown, col. 3, line 59 – col. 4, line 15; col. 4, lines 12-15.; col. 7, lines 55-65; col. 9, lines 5-10, col. 10, line 63 - col. 11, line 28).

The Examiner acknowledged that Brown is missing the step of identifying an error dispenser. Answer at 5. To address this deficiency, the Examiner looks to Tisone, which discloses determination of a phase adjustment by the controller for each dispense cycle either before or during production such that a high degree of accuracy, precision, and repeatability is attained. Answer at 5 (citing Tisone, col. 8, lines 48-55).

The Examiner concludes that it would have been obvious to a person of ordinary skill in the art at the time of the invention to include the step of identifying an error dispenser as taught by Tisone in the method of Brown. Answer at 5. Moreover, the Examiner states one of ordinary skill in the art would have been motivated to include the step of identifying an error dispenser in the method of Brown for the advantage of providing an apparatus dispenser system with a control system that precisely coordinates dispensing operations with a high degree of accuracy, precision, and repeatability. *Id.* at 5.

Furthermore, in response to Appellants' argument that neither Brown nor Tisone teaches or suggests at least one set of redundant dispensers in each group of dispensers and claimed steps 1(c) and 1(d), the Examiner maintains that Brown and Tisone suggest the limitations of claim 1 because a group of dispensers can be a single row of dispensers and a set can be just one dispenser. *Id.* at 10-11. The Examiner supports their argument by citing the specification's definition of a "set" or "sub-set" of any item, which can include just one of the items. *Id.* at 11 (citing Specification at 8).

We find the Examiner's position to be inconsistent with the claim language. "It is a 'bedrock principle' of patent law that 'the claims of a patent define the invention to

which the patentee is entitled the right to exclude." *Phillips v. AWH Corp.*, 415 F.3d 1303, 1312, 75 USPQ2d 1321, 1325 (Fed. Cir. 2005) (citations omitted). "Quite apart from the written description and the prosecution history, the claims themselves provide substantial guidance as to the meaning of particular claim terms." *Id.* at 1314, 75 USPQ2d at 1327 (citations omitted). For example, "the use of a term within the claim provides a firm basis for construing the term." *Id.* Additionally, claims "must be read in view of the specification, of which they are a part." *Id.* at 1315, 75 USPQ2d at 1327. "Of course, at all times, the language of the claims governs their scope and meaning" and "[u]nless the intrinsic evidence compels a contrary conclusion, the claim language carries the meaning accorded those words in the usage of skilled artisans at the time of invention." *Smithkline Beecham Corp. v. Apotex Corp.*, 403 F.3d 1331, 1338-39, 74 USPQ2d 1396, 1403 (Fed. Cir. 2005).

The Examiner has taken the claim term "set" out of the context of the claim. Here, claim 1 reads in part:

- Claim element 1(a): the step of "loading the dispensers with fluid such that each dispenser group has at least one *set of redundant dispensers* loaded with a same fluid;"
- Claim element 1(c): the step of "moving a first dispenser of each set in each group along the selected path for that group while dispensing drops from non-error first dispensers of the sets in at least part of the pattern along the selected path for each group;" and
- Claim element 1(d): the step of "moving *a second dispenser* of the sets in each group along the selected path for that group while dispensing drops from *a non-error second dispenser of a set having an identified error first dispenser*, in at least part of the pattern for the selected path of the first group."

(Emphasis added). This claim language makes clear that it *requires* a dispenser group with at least one "set of *redundant* dispensers." The specification states that dispensers within a set are "redundant in that one can be used *in place of the other* during array fabrication (assuming the one used in place is functioning and is not in error in some way)." Specification at 12 (emphasis added). Redundancy of dispensers necessarily requires at least one dispenser *redundant to another*. The specification defines a "set" or "subset" of any item as containing "only one of the item, or only two, or three, or any number of multiple items." Specification at 8. Thus, we find that a "set of redundant dispensers" can be one, two, or three, or any number of dispensers *redundant to another.*

We agree with Appellants that "a '*set*' of dispensers as recited in Claim 1 **must** have more than one dispenser to perform the claimed methods" and this limitation is clear from reading the claims. Reply at 2. As indicated by Appellants, the "only way that [a second dispenser in a set could dispense drops where error dispensers did not] is for a *set* of dispensers to have more than one dispenser" and "a *set* of dispensers as claimed in Claim 1 **must** have at least two dispensers." Reply at 3.

Claim 1

After careful review of the Brown and Tisone patents, we find that the combined or modified teachings of the references fail to teach a method of error identification and correction that utilizes redundant dispensers. Brown does not add anything to the Examiner's argument for prima facie obviousness. Moreover, although Tisone teaches a head system with multiple groups of drop dispensers containing the same fluid, arranged in a coordinated fashion, and discloses error identification through experimentation,

Tisone fails to teach a method that utilizes the multiple, redundant dispensers for error correction as recited in claim elements 1(c-d), 6(c-d) and 25(c-d).

Rather, we agree with Appellants that

> the error identification method disclosed in Tisone et al. is directed to controlling specific parameters of valve deposition (e.g., timing) and does not teach identifying an error dispenser and dispensing only from non-error dispensers during array fabrication as is claimed. Instead, the deposition error method of Tisone et al. evaluates whether a drop is deposited in the desired location and, if it is not, adjustments are made to the parameters of valve deposition to correct it. In other words, there is no such thing as an "error dispenser" in Tisone et al. as claimed in the subject application. If a dispenser deposits erroneously, the method disclosed in Tisone et al. adjusts the parameters of dispensation and deposits fluid using the *same* dispenser.

Brief at 14.

Claims 2-33

Claims 2-33 all require redundant dispensers. Thus, we reverse the rejection of these claims for the reasons we reverse the rejection of claim 1. See our analysis *supra* at pp. 11-16.

Although the Examiner separately rejected dependent claims 4, 20, and 30 under § 103(a) as being unpatentable over Brown, Tisone, and Gamble, we reverse the rejection of these claims without further analysis. Gamble does not disclose or suggest redundant dispensers as claimed and therefore does not resolve the deficiencies of the § 103(a) rejection of the relevant independent claims (1, 6, and 25) from which claims 4, 20,and 30 depend, respectively. The Examiner's rejections for claims 2-33 are reversed.

Other issues

The following printer technology prior art references may be reasonably pertinent to the inventors' use of redundant dispensers to correct for error dispensers:

- Kumar et al., U.S. Patent No. 6,283,572 issued Sept. 4, 2001, for "Dynamic Multi-Pass Print Mode Corrections to Compensate for Malfunctioning Inkjet Nozzles" (Figure 7, Tables I and II, in particular);
- Anderson, U.S. Patent No. 6,076,910 issued June 20, 2000, for "Ink Jet Printing Apparatus having Redundant Nozzles" (claim 13, in particular); and
- Hackleman, U.S. Patent No. 5,640,183 issued June 17, 1997, for "Redundant Nozzle Dot matrix Printheads and Method of Use."

It would have been reasonable for one skilled in the art to look to the field of printer technology at the time of invention given that microarray production companies were innovating with concepts borrowed from printer technology as early as 1998:

- Industrial Technology Research Institute News Release "Implementation of Phalanx Microarray Technology—Fruition of ITRI's Multidisciplinary Effort in Biotechnology," http://www.itri.org.tw/eng/news/spotlight-show.jsp?path=f-20030409.dcr;
- ArrayJet History, http://www.arrayjet.co.uk/about.html;
- MacBeath, "Printing Proteins as Microarrays for High-Throughput Function Determination," *Science*, New Series, col. 289, No. 5485 (Sep. 8, 2000), pp. 1760-1763; and
- Shimadzu Biotech Press Release October 2001, "Proteome Systems and Shimadzu Biotech Complete 1st Stage of the Chemical Printer Development," http://www.shimadzu-biotech.net/pages/news/1/press_releases/2001_10_a_proteome.php.

(Copies of the internet articles are included in the Appendix.) The Examiner should consider these references before the application is allowed to issue as a patent.

REVERSED

DONALD E. ADAMS
Administrative Patent Judge

DEMETRA J. MILLS
Administrative Patent Judge

NANCY J. LINCK
Administrative Patent Judge

BOARD OF PATENT APPEALS AND INTERFERENCES

APPENDIX

Notice of References Cited	Application/Control No. 10/061,800	Applicant(s)/Patent Under Reexamination 2006-2664	
	Examiner BPAI	Art Unit 1600	Page of

U.S. PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Name	Classification
	A	US- 6,283,572		Kumar et. al.	
	B	US- 6,076,910		Anderson	
	C	US- 5,640,183		Hackleman	
	D	US-			
	E	US-			
	F	US-			
	G	US-			
	H	US-			
	I	US-			
	J	US-			
	K	US-			
	L	US-			
	M	US-			

FOREIGN PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Country	Name	Classification
	N					
	O					
	P					
	Q					
	R					
	S					
	T					

NON-PATENT DOCUMENTS

*		Include as applicable: Author, Title Date, Publisher, Edition or Volume, Pertinent Pages)
	U	Industrial Technology Research Institute News Release "Implementation of Phalanx Microarray Technology, "Fruition of ITRI's Multidisciplinary Efforts in Biotechnology," http://www.itri.org.tw/eng/news/spotlight-show.jsp?path=f-20030409.dcr;
	V	Array Jet History, http://www.arrayjet.co.uk/about.html;
	W	
	X	

*A copy of this reference is not being furnished with this Office action. (See MPEP § 707.05(a).)
Dates in MM-YYYY format are publication dates. Classifications may be US or foreign.

Implementation of Phalanx Microarray Technology-

- Fruition of ITRI's Multidisciplinary Effort in Biotechnology

Background

The research field of molecular biology began with the discovery of DNA structure in 1953, and has gained a great wealth of knowledge and revolutionized the study of biology and medicine. The more application-oriented side is biotechnology, which in recent years has caught the world's attention as a steady stream of success stories on cloning, genetic modification and human genome project appears in the news. Researchers in biotechnology are continuously searching for devices and methods with better precision and higher throughput.

DNA Microarrays

With the availability of whole genome sequences, many tools were developed to study biology on a whole-genome scale. The DNA Microarray, or Gene Chip, is the most important invention of them all. A DNA microarray is a slide (a few square centimeters) that contains many different kinds of DNA (called "probes") deposited on its surface, each based on a certain gene from a genome of interest. The probes on the slide usually are arranged in an array, each address (or spot) on the DNA microarray corresponds to a specific gene. The probe on the slide can grab or "hybridize with" the complementary DNA or RNA fragments (called "targets") generated from the testing sample. By measuring the fluorescent intensity on a probe location after hybridization, one can estimate the expression activity of a specific gene in the testing sample.

Manufacturing Processes for DNA Microarrays

Currently there are two basic ways of making DNA microarrays: the in-situ

synthesis and the spotting methods.
The in-situ method performs the direct synthesis of DNA molecules on the surface of the microarray slide. Tens of thousands of DNA synthesis reactions are carried out simultaneously on the slide surface. There are two different in-situ synthesis methods, i.e., photolithography and inkjet printing. The photolithography method borrows technology developed in the semi-conductor industry. A series of specially designed photo-masks are used to introduce in sequence the photoactive analogs of the four DNA nucleotides (A, C, T, and G) into the synthesis reactions. The other in-situ method uses the inkjet printing mechanism to deliver the DNA nucleotides onto the probe location. The inkjet-head movement is computer-controlled to ensure the accuracy of the nucleotide deposition process. For both methods, the quality of the DNA is very difficult to monitor or control. They also suffer from high manufacturing cost and low production capacity. The unit price ranges from US$500 to US$2,000.
In the spotting method, the probes are synthesized before they are applied to the microarray surface. The probe is usually synthesized by polymerase chain reaction (for the longer cDNA probe) or by a conventional DNA synthesis method. The probes are then spotted on the slide and immobilized through various surface chemistry mechanisms. The effectiveness of this method is highly dependent on the design of the arraying equipment and the surface chemistry between the probe solution, the dispensing apparatus, and the slide surface.
Currently there are many robotic microarrayers and microarray slides available on the market for smaller scale production. The systems are usually set up by the microarray core facility of research institutes for in-house usage. The throughput and the production size are relatively low, so the unit cost stays high. The quality of the microarrays is inconsistent, making comparison between various microarray experiments very difficult if not impossible. ITRI has now come up with a manufacturing scheme that combines the advantages of in-situ and spotting methods, resulting in significantly higher throughput and lower cost.

ITRI's Phalanx Microarray Technology - a High Throughput Manufacturing Process

In 1998, theBiomedical Engineering Center (BMEC)of ITRI initiated the Biochip Project to explore the potential of microarray technology. The multidisciplinary research team of the project came from 5 different research ITRI divisions, including BMEC,Opto-Electronics & Systems Laboratories (OES),Center for Measurement Standards (CMS),Union Chemical Laboratories(UCL), andElectronics Research & Service Organization (ERSO). The project has led to a multitude of patents, covering the subjects of surface chemistry, microdispenser, microarray, and electrophoresis. The collective result of the project is the phalanx microarray technology, which is a high throughput manufacturing process that can produce reliable, high-quality microarrays with a density of 4,000 pre-synthesized probes per cm2 at low cost, perhaps as low as one-tenth of that of the current product

The core of the phalanx microarray technology is the Phalanx Jet liquid micro-dispenser, Phalanx Arrayer, and Phalanx Slide. The Phalanx Jet and Phalanx Array were co-developed by BMEC and OES. The Phalanx Jet

employs bubble jet printer technology to precisely dispense micro-volume liquid at very high density. The Phalanx Arrayer is an automatic arraying platform that can be assembled into a continuous arraying pipeline with high precision and throughput. BMEC and UCL co-developed the surface chemistry for the Phalanx Slide that enables the DNA solution to maintain a uniform contact surface and to maximize the DNA immobilization on the slide surface.

The Founding of Phalanx Biotechnology Group, Inc.

Due to the great success of the Biochip Project, ITRI and other local biotech businesses formed the Biochip R&D Alliance to pursue the accompanying commercial opportunities. To make the best use of the Project's IP, ITRI put together an IP bundle and licensed it exclusively to the new start-up formed by that Alliance. The Alliance has invested an aggregate of 500 million NT dollars to create Phalanx Biotech Group, Inc. (PBG) to implement the micro-array production technology, staffed mainly by members from the Project.

PBG will have a pilot product Phalanx Human Liver 2000 Microarray, which contains about 2000 probes for liver related genes, by April 2003. It will begin producing Phalanx Human Whole-Genome Microarray (PHWGM), containing more than 30,000 probes that cover all known genes in human genome, by the end of 2003. PBG will design all the probe sequences collaboratively with ITRI using BMEC's bioinformatics software, which will incorporate the most updated human genome information. PBG will also continue to work with BMEC to adopt state-of-the-art quality control concepts into microarray production, such as using MALDI-TOF mass spectrometry to validate the integrity and identity of every probe on the microarray. Furthermore, PBG will continue to work in partnership with other ITRI divisions for the improvement in phalanx microarray technology.

Related Link: http://www.bmec.itri.org.tw/english/main.htm

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Last Updated: 08/31/2006

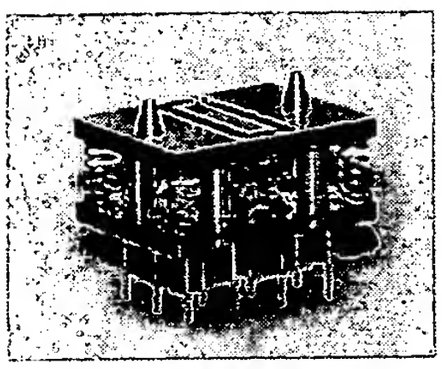
Aj Arrayjet

High Quality Microarray Production Powered by Ink

History

Arrayjet ® Ltd was founded in August 2000 by a Cambridge physicist, Dr. Howard Mann of Edinburgh molecular biologists, Prof. Peter Ghazal and Dr. Douglas Roy, to develop ro printheads to make biological microarrays. In February 2001 Arrayjet secured two-stag Scotland based investor group known as Archangels and won a Scottish Enterprise develop the ink jet microarray platform. Further funding by Archangels and SE followed.

Dr. Manning is Arrayjet's Technical Director and Keith Howell, an Edinburgh based direct is Chairman. Professor Ghazal is chief scientific advisor and Dr Roy is lead advisor on engineers have been recruited, premises located close to Edinburgh and a labora professional alliances have been formed.



Arrayjet was at first engaged in product development. In the period from February 200 year the fundamental technology behind Arrayjet was demonstrated, and milestones sel the first round of funding were met. Subsequent funding supported the design, commissioning of a pre-production prototype.

Arrayjet has now launched a number of products into the microarray market and contin technology.

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as single-nucleotide polymorphism analysis, where single-mismatch resolution, sensitivity, cost, and ease of use are important factors. Moreover, the sensitivity of this system, which has yet to be totally optimized, points toward a potential method for detecting oligonucleotide targets without the need for target amplification schemes such as the polymerase chain reaction.

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10. For the experiments reported in Fig. 2, dissociation measurements were made from the surface of glass beads 250 to 300 μm in diameter (Polysciences, Warrington, PA) rather than planar substrates to increase the UV-visible and fluorescence signal intensity.
11. 5′-Cy3–labeled oligonucleotide probes were synthesized on an Expedite automated synthesizer (Millipore, Bedford, MA) using Cy3 phosphoramidite (Glen Research, Sterling, VA) as the label source. Arrays of spots 175 μm in diameter separated by 375 μm were patterned with a GMS 417 Microarrayer (Genetic Microsystems, Woburn, MA).
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18. C.A.M. and R.L.L. acknowledge the Army Research Office (DAAGS5-97-1-0133) and the National Institute of General Medical Sciences (GM 57356) for support of this work.

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REPORTS

Printing Proteins as Microarrays for High-Throughput Function Determination

Gavin MacBeath[1]* and Stuart L. Schreiber[2]

Systematic efforts are currently under way to construct defined sets of cloned genes for high-throughput expression and purification of recombinant proteins. To facilitate subsequent studies of protein function, we have developed miniaturized assays that accommodate extremely low sample volumes and enable the rapid, simultaneous processing of thousands of proteins. A high-precision robot designed to manufacture complementary DNA microarrays was used to spot proteins onto chemically derivatized glass slides at extremely high spatial densities. The proteins attached covalently to the slide surface yet retained their ability to interact specifically with other proteins, or with small molecules, in solution. Three applications for protein microarrays were demonstrated: screening for protein-protein interactions, identifying the substrates of protein kinases, and identifying the protein targets of small molecules.

Historically, genome-wide screens for protein function have been carried out with random cDNA libraries. Most frequently, the libraries are prepared in phage vectors and the expressed proteins immobilized on a membrane by a plaque lift procedure. This method has been effective for a variety of applications (*1*–*4*), but it has several limitations. Most clones in the library do not encode proteins in the correct reading frame, and most proteins are not full-length. Bacterial expression of eukaryotic genes frequently fails to yield correctly folded proteins, and products derived from abundant transcripts are overrepresented. Moreover, because plaque lifts are not amenable to miniaturization on the micrometer scale, it is hard to imagine screening all the proteins of an organism hundreds or thousands of times by this approach.

With the advent of high-throughput molecular biology, it is now possible to prepare large, normalized collections of cloned genes. UniGene sets in the form of polymerase chain reaction products have been used extensively over the past decade to construct DNA microarrays for the study of transcriptional regulation (*5*). Recently, spatially segregated clones in expression vectors were used to study protein function in vivo using the yeast two-hybrid system (*6*) and in vitro using biochemical assays (*7*). We have built on these efforts by developing microarray-based methods to study protein function.

To accomplish these goals, it is necessary to immobilize proteins on a solid support in a way that preserves their folded conformations. One group has described methods of arraying functionally active proteins, using microfabricated polyacrylamide gel pads to capture their samples and microelectrophoresis to accelerate diffusion (*8*). In contrast, we have immobilized proteins by covalently attaching them to the smooth, flat surface of glass microscope slides. One of our primary objectives in pursuing this approach was to make the technology easily accessible and compatible with standard instrumentation. We use a variety of chemically derivatized slides that can be printed and imaged by commercially available arrayers and scanners. For most applications, we use slides that have been treated with an aldehyde-containing silane reagent (*9*). The aldehydes react readily with primary amines on the proteins to form a Schiff's base linkage. Because typical proteins display many lysines on their surfaces as well as the generally more reactive α-amine at their NH_2-termini, they can attach to the slide in a variety of orientations, permitting different sides of the protein to interact with other proteins or small molecules in solution.

To fabricate protein microarrays, we use a high-precision contact-printing robot (*10*) to deliver nanoliter volumes of protein samples to the slides, yielding spots about 150 to 200 μm in diameter (1600 spots per square centimeter). The proteins are printed in phosphate-buffered saline with 40% glycerol included to prevent evaporation of the nanodroplets. It is important that the proteins remain hydrated throughout this and subsequent steps to prevent denaturation. After a 3-hour incubation, the slides are immersed in a buffer containing bovine serum albumin (BSA). This step not only quenches the unreacted aldehydes on the slide, but also forms a molecular layer of BSA that reduces nonspecific binding of other proteins in subsequent steps.

Although appropriate for most applications, aldehyde slides cannot be used when peptides

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or very small proteins are printed, presumably because the BSA obscures the molecules of interest. For such applications, we use BSA-NHS (BSA-*N*-hydroxysuccinimide) slides that are fabricated by first attaching a molecular layer of BSA to the surface of glass slides and then activating the BSA with *N,N'*-disuccinimidyl carbonate (*11*). The activated lysine, aspartate, and glutamate residues on the BSA react readily with surface amines on the printed proteins to form covalent urea or amide linkages. The slides are then quenched with glycine. In contrast to the aldehyde slides, proteins or peptides printed on BSA-NHS slides are displayed on top of the BSA monolayer, rendering them accessible to macromolecules in solution.

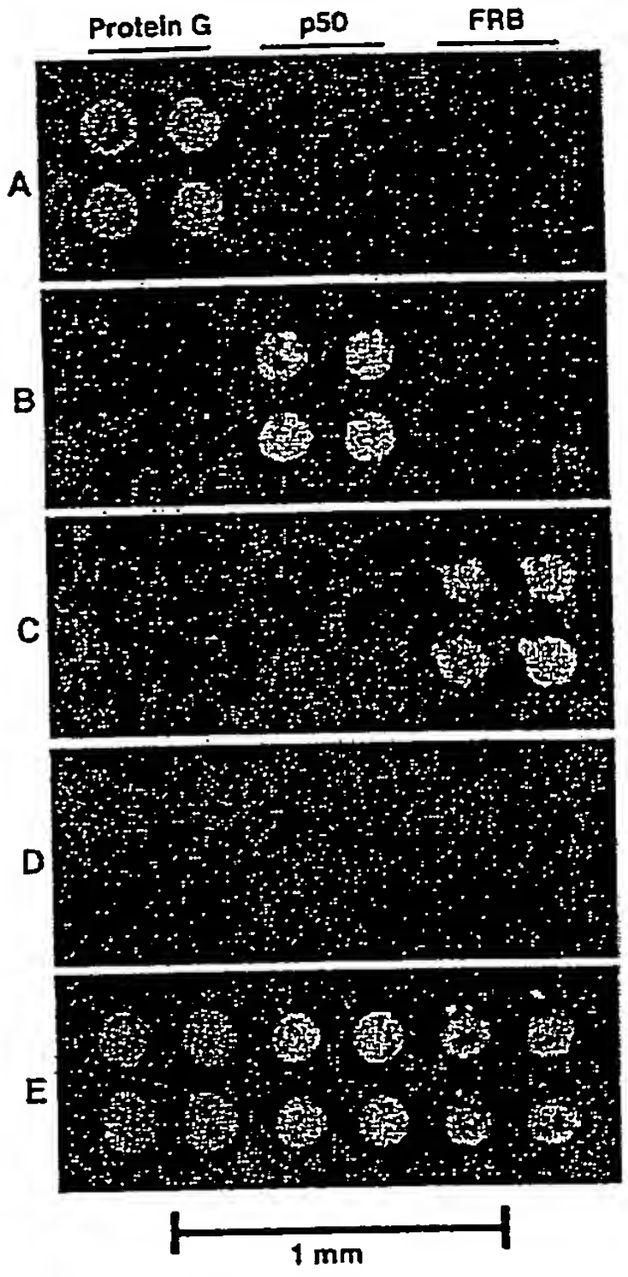


Fig. 1. Detecting protein-protein interactions on glass slides. (A) Slide probed with BODIPY-FL-IgG (0.5 μg/ml). (B) Slide probed with Cy3-IκBα (0.1 μg/ml). (C) Slide probed with Cy5-FKBP12 (0.5 μg/ml) and 100 nM rapamycin. (D) Slide probed with Cy5-FKBP12 (0.5 μg/ml) and no rapamycin. (E) Slide probed with BODIPY-FL-IgG (0.5 μg/ml), Cy3-IκBα (0.1 μg/ml), Cy5-FKBP12 (0.5 μg/ml), and 100 nM rapamycin. In all panels, BODIPY-FL, Cy3, and Cy5 fluorescence were false-colored blue, green, and red, respectively.

As a first application of protein microarrays, we have looked at protein-protein interactions. Until now, only the yeast two-hybrid system has been used to investigate such interactions systematically on a genome-wide scale (*6*). This in vivo method, although easy to implement and of great utility, has several limitations. Proteins that function as transcriptional activators yield false positives when expressed as DNA binding domain fusions. False negatives are encountered when proteins are displayed inappropriately or when the DNA binding domain fusions are produced in excess. Proteins that do not fold correctly in yeast are inaccessible, and posttranslational modifications (such as phosphorylation or glycosylation) cannot be controlled. Finally, it is impossible to control the environment (e.g., ion concentration, presence or absence of cofactors, temperature) during the experiment.

To determine whether microarrays could be used for these types of studies, we selected three pairs of proteins that are known to interact: protein G and immunoglobulin G (IgG) (*12*); p50 (of the nuclear factor NF-κB complex) and the NF-κB inhibitor IκBα (*13*); and the FKBP12-rapamycin binding (FRB) domain of FKBP-rapamycin–associated protein (FRAP) and the human immunophilin FKBP12 (12 kD FK506-binding protein) (*14*). The first two interactions occur without special requirements, whereas the third interaction depends on the presence of the small molecule rapamycin (*14*). We arrayed the first protein of each pair in quadruplicate on five aldehyde slides and probed each slide with a different fluorescently labeled protein (*11*).

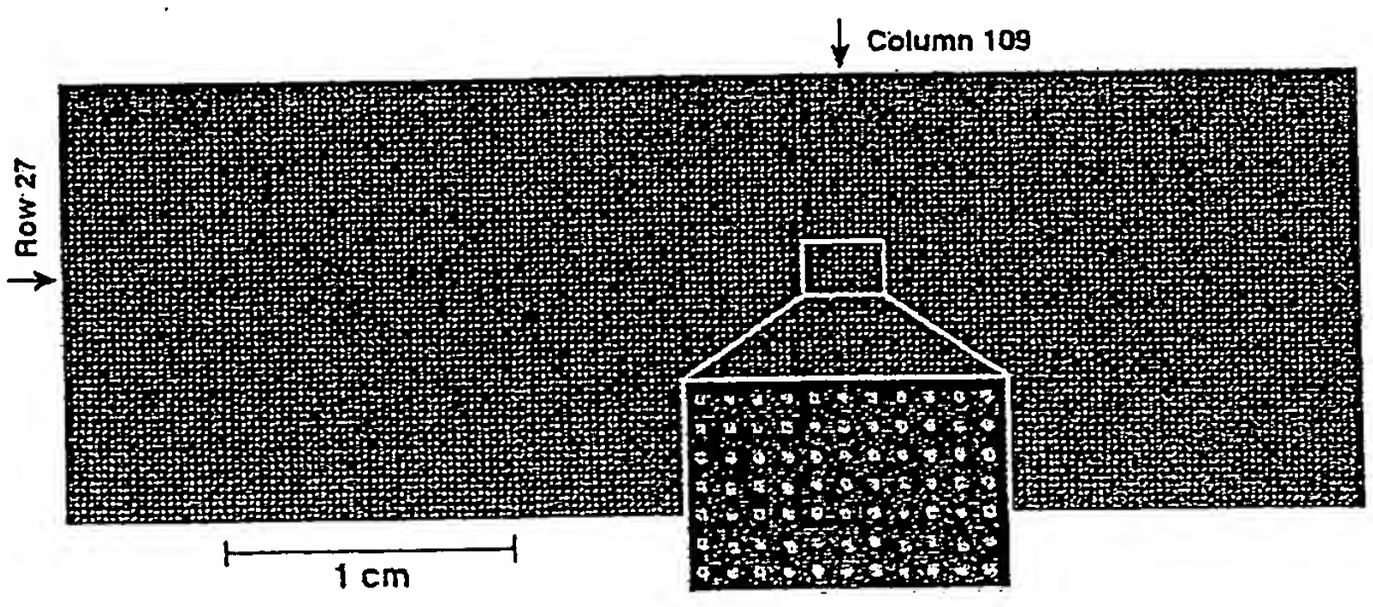
The slide in Fig. 1A was probed with BODIPY-FL–conjugated IgG, washed, and scanned with an ArrayWoRx fluorescence slide scanner (*15*). As anticipated, only the spots containing protein G were visible, indicating that the immobilized protein is able to retain its functional properties on the glass surface. Similarly, only the p50-containing spots were visible on the slide probed with Cy3-IκBα (Fig. 1B) (*15*). For Cy5-FKBP12, binding to FRB was observed only when rapamycin was added (Fig. 1, C and D). Because the three fluorophores used for these studies have nonoverlapping excitation and emission spectra, we were also able to detect these interactions simultaneously (Fig. 1E).

By varying the concentration of FRB (the protein being immobilized), we found that at concentrations above 1 mg/ml, the fluorescence of the spots began to saturate. Below this, fluorescence scaled linearly with decreasing concentrations of FRB. All proteins immobilized on the slides described here were spotted at 100 μg/ml. Because only a few microliters of each protein are sufficient to fabricate thousands of microarrays, purified proteins may be readily obtained by high-throughput expression and purification, or even by in vitro transcription/translation (*16*).

Much lower concentrations are needed for the solution-phase protein. In the case of Cy5-FKBP12, fluorescence scaled linearly with protein concentration over four orders of magnitude (*11*). Specific binding could be detected using Cy5-FKBP12 concentrations as low as 150 pg/ml (~12.5 pM). Concentrations in this range are accessible not only with purified proteins, but also with fluorescently labeled proteins from cell lysates. Thus, specific interactions, once defined, may potentially be exploited to quantify protein abundance and modification in whole cells or tissues.

At the spot density used for these studies, it was possible to fit more than 10,000 samples in about half the area of a standard (2.5 cm by 7.5 cm) slide. To investigate the feasibility of detecting a single specific interaction in this larger context, we prepared a slide containing 60 rows and 180 columns of spatially separated spots. Protein G was spotted 10,799

Fig. 2. A single slide holding 10,800 spots. Protein G was printed 10,799 times. A single spot of FRB was printed in row 27, column 109. The slide was probed with BODIPY-FL-IgG (0.5 μg/ml), Cy5-FKBP12 (0.5 μg/ml), and 100 nM rapamycin. BODIPY-FL and Cy5 fluorescence were false-colored blue and red, respectively.



times on this slide, with a single spot of FRB in row 27, column 109. The slide was then probed with a mixture of BODIPY-FL-IgG and Cy5-FKBP12, with 100 nM rapamycin included in the buffer. Figure 2 shows the single FRB spot, clearly visible in the sea of protein G spots.

Although it is of great value to identify stable protein-protein interactions in a system such as a cell or tissue, it is equally important to define the transient interactions that occur between enzymes and their substrates. Protein microarrays offer an ideal system, for example, for the rapid and parallel identification of the substrates of protein kinases. To investigate this application, we chose three different kinase-substrate pairs: adenosine 3′,5′-monophosphate–dependent protein kinase (PKA) and Kemptide (a peptide substrate for PKA) (*17*); casein kinase II (CKII) and protein phosphatase inhibitor 2 (I-2) (*18*); and p42 mitogen-activated protein (MAP) kinase (Erk2) and Elk1 (*19*). The protein substrates of each pair were spotted in quadruplicate onto three BSA-NHS slides, and each slide was incubated with a different kinase in the presence of [γ-^{33}P]adenosine triphosphate.

Although isotopic labeling of the protein spots is the most direct way to identify phosphorylation, the challenge lies in detecting the radioactive decay. Neither x-ray film nor conventional PhosphorImagers offer sufficient spatial resolution to visualize the spots, which are 150 to 200 μm in diameter. Borrowing from the technique of isotopic in situ hybridization, we dipped the slides in a photographic emulsion and developed them manually; this resulted in the deposition of silver grains directly on the glass surface. The slides were then visualized using an automated light microscope (*20*) and individual frames were stitched together. As anticipated, only the specific substrates for each enzyme were phosphorylated (Fig. 3).

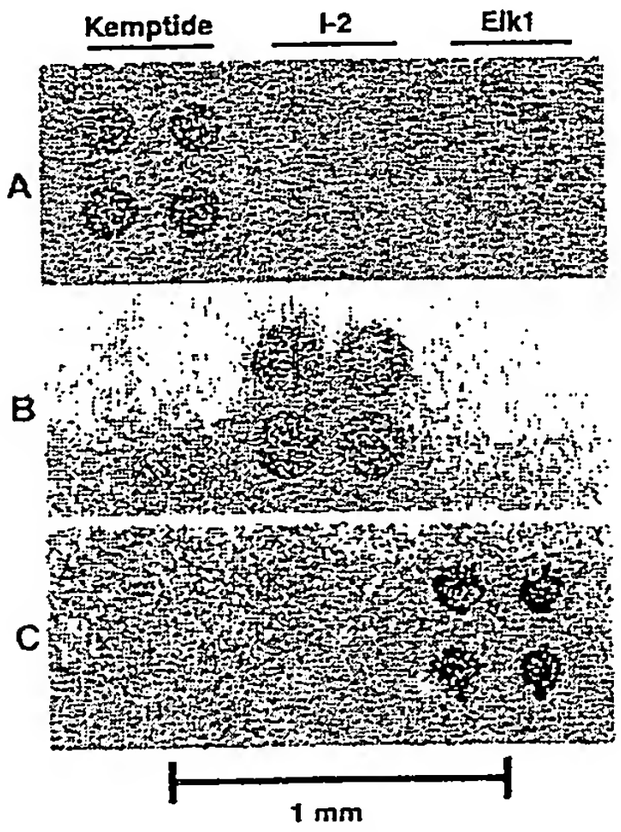


Fig. 3. Detecting the substrates of protein kinases on glass slides. (A) Slide incubated with the catalytic subunit of PKA. (B) Slide incubated with CKII. (C) Slide incubated with p42 MAP kinase (Erk2).

As the third and most demanding application, we sought to use protein microarrays to identify protein–small molecule interactions. With the advent of high-throughput, cell-based screening, more and more compounds are being identified on the basis of their biological activity. Once a "hit" is obtained, the daunting task of target identification remains. Several innovative techniques have been developed to address this bottleneck (*4*, *21*–*23*), but they all suffer from the common limitations imposed by using random cDNA libraries. As an alternative, we sought to develop microarray-based assays that use purified, full-length, correctly folded proteins.

To test this approach, we chose three unrelated small molecules for which specific protein receptors are available: DIG, a derivative of the steroid digoxigenin that is recognized by a mouse monoclonal antibody (*24*); biotin, a common vitamin recognized by the bacterial protein streptavidin (*25*); and AP1497 (Fig. 4), a synthetic pipecolyl α-ketoamide designed to be recognized by FKBP12 (*26*). The proteins from all three pairs were spotted in quadruplicate on four aldehyde slides, and each slide was probed with a different small molecule. Rather than labeling the compounds directly, each ligand was coupled to BSA that had previously been labeled with a unique fluorophore ($Alexa_{488}$, Cy3, or Cy5) (*15*). As anticipated, fluorescence localized to the appropriate spots in all three cases (Fig. 5, A to C). Because the fluorophores used for these studies have non-overlapping excitation and emission spectra, we were also able to detect all three interactions simultaneously (Fig. 5D).

To investigate our ability to detect low-affinity interactions, we prepared Cy3-BSA conjugates of compounds AP1497, AP1767, and AP1780 (Fig. 4; dissociation constants for FKBP12 of 8.8 nM, 140 nM, and 2.6 μM, respectively). When three identical slides displaying FKBP12 were probed in parallel, spots with comparable fluorescence intensities were obtained for all three conjugates (*11*). This means that interactions in the micromolar range can easily be observed. The fact that the intensity of the fluorescence did not vary appreciably as the affinity of the interaction was lowered can be attributed to the multivalency of the BSA conjugates (avidity effects). In the context of small-molecule microarrays (*27*), we have previously shown that when these three compounds are immobilized on a glass surface and then probed with Cy5-labeled FKBP12 (a monomeric protein), the intensity of the fluorescence correlates very well with the affinity of the interaction. Thus, by controlling the valency of the probe, we can choose whether to observe differences in affinity or to favor the detection of low-affinity interactions. The combination of these two approaches may prove useful in the identification of both primary and secondary drug targets.

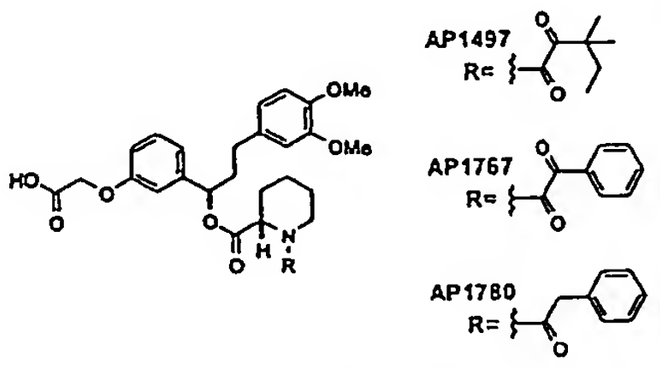


Fig. 4. Synthetic ligands for FKBP12. The compounds were coupled to BSA through their carboxyl groups (via a flexible linker).

Although traditional biochemical methods have yielded invaluable insight into protein function on a case-by-case basis, they cannot realistically be applied to the study of every protein in a cell, tissue, or organism. If we hope to assign function on a broader level, we must turn to miniaturized assays that can be performed in a highly parallel format. It is certainly a daunting task to express and purify thousands of different proteins, and some

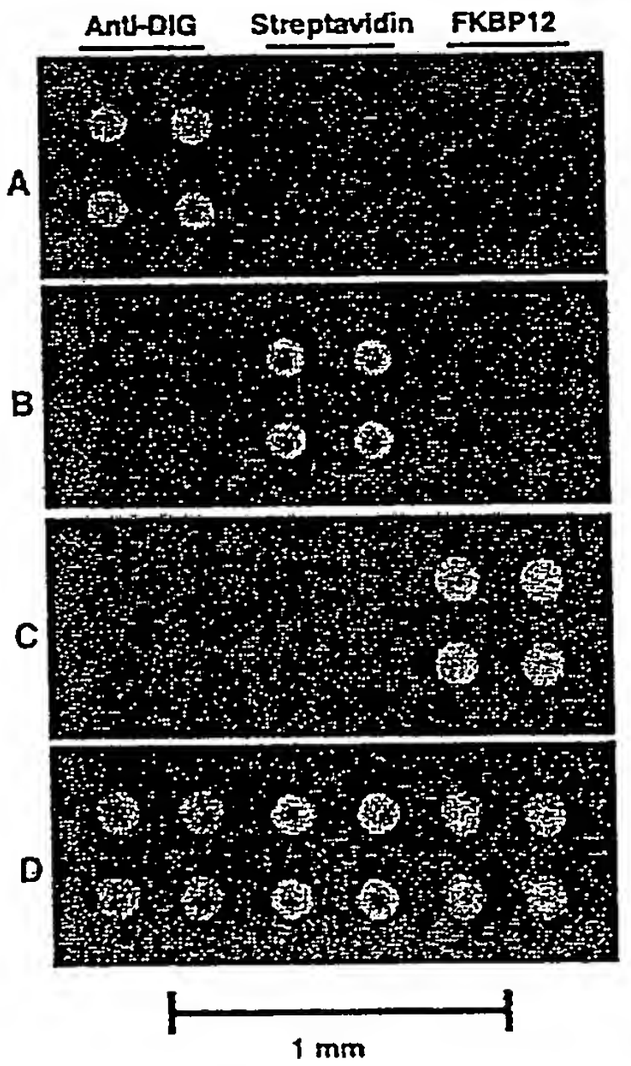


Fig. 5. Detecting the targets of small molecules on glass slides. (A) Slide probed with $Alexa_{488}$-BSA-DIG. (B) Slide probed with Cy5-BSA-biotin. (C) Slide probed with Cy3-BSA-AP1497. (D) Slide probed with $Alexa_{488}$-BSA-DIG, Cy5-BSA-biotin, and Cy3-BSA-AP1497. All conjugates were used at a concentration of 10 μg/mL In all panels, $Alexa_{488}$, Cy3, and Cy5 fluorescence were false-colored blue, green, and red, respectively.

proteins will inevitably prove refractory to biochemical manipulation. Nonetheless, the effort will be worthwhile if the many proteins that are amenable can be assayed both simultaneously and repeatedly. By fabricating protein microarrays, we can fulfill both these criteria, facilitating the in vitro study of protein function on a genome-wide level.

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28. We thank R. Peters and T. Maniatis at Harvard University for samples of p50 and IκBα and D. Holt and T. Clackson at Ariad Pharmaceuticals Inc. for samples of AP1497, AP1767, and AP1780. We thank the Harvard Center for Genomics Research for support of the G.M. laboratory and the National Institute of General Medical Sciences for support of the S.L.S. laboratory. G.M. was also supported in part by a fellowship from the Cancer Research Institute. S.L.S. is an HHMI investigator.

19 June 2000; accepted 14 July 2000

REPORTS

The Global Spread of Malaria in a Future, Warmer World

David J. Rogers[1]* and Sarah E. Randolph[2]

The frequent warnings that global climate change will allow *falciparum* malaria to spread into northern latitudes, including Europe and large parts of the United States, are based on biological transmission models driven principally by temperature. These models were assessed for their value in predicting present, and therefore future, malaria distribution. In an alternative statistical approach, the recorded present-day global distribution of *falciparum* malaria was used to establish the current multivariate climatic constraints. These results were applied to future climate scenarios to predict future distributions, which showed remarkably few changes, even under the most extreme scenarios.

Predictions of global climate change have stimulated forecasts that vector-borne diseases will spread into regions that are at present too cool for their persistence (*1*–*5*). For example, life-threatening cerebral malaria, caused by *Plasmodium falciparum* transmitted by anopheline mosquitoes, is predicted to reach the central or northern regions of Europe and large parts of North America (*2*, *4*). *falciparum* malaria is the most severe form of the human disease, causing most of the ~1 million deaths worldwide among the ~273 million cases in 1998 (*6*). Despite these figures, the epidemiology of malaria, like many other vector-borne tropical diseases, remains inadequately understood. Only the most general of maps for its worldwide distribution are available (*7*), and its global transmission patterns cannot be modeled satisfactorily because crucial parameters and their relations with environmental factors have not yet been quantified. Most importantly, absolute mosquito abundance has not yet been related to multivariate climate.

Nevertheless, the problem of malaria has led to its being included in most predictions about the impact of climate change on the future distribution of vector-borne diseases (*8*). These studies, which draw on the forecasts of future climate from various global circulation models (GCMs) (*9*, *10*), generally use only one or at most two climatic variables to make their predictions. Biological models for malaria distribution are based principally on the temperature dependence of mosquito blood-feeding intervals and longevity and the development period of the malaria parasite within the mosquito, each of which affects the rate of transmission (*4*, *11*). Those models based on threshold values include a lower temperature threshold, below which all development of the malaria parasite ceases, and an upper limit of mosquito lethality (*2*). In addition, the suitability (or unsuitability) of habitats for these vectors, which require a minimum atmospheric moisture, is defined by the ratio of rainfall to potential evapotranspiration (*2*). The output of such models, therefore, represents predicted areas where parasite development within the vector is fast enough to be completed before the vector dies, bounded by limits imposed by habitat suitability (*2*). The fit of these predictions to the current global malaria situation shows noticeable mismatches in certain places (*12*); false predictions of presence (e.g., over the eastern half of the United States) are accounted for by past control measures or by "peculiar vector biogeography," whereas false predictions of absence are dismissed as model errors (*2*).

Refinements of these biological models (*3*–*5*) are based on modifications of an equation describing transmission potential, expressed as the basic reproduction number R_0, which must equal at least 1 for disease persistence (*13*, *14*). For an estimation of the correct value of R_0 from which to predict malaria distribution, absolute, not relative, estimates of all quantities in the equation are needed. Instead, by omitting certain unquantified but important parameters and rearranging the equation (*15*), a relative measure of "epidemic potential" (EP) [now "transmission potential" (*5*)] has been derived as the reciprocal of the vector/host ratio required for disease persistence. This predicts a more extensive present-day distribution of malaria than is currently observed (*12*). The ratio of future EP to present EP is then presented as indicating the relative degree of the future risk of malaria, but this is an inappropriate measure of changing risk because a high ratio may still leave $R_0 < 1$.

Until such biological approaches can give accurate descriptions of the current situation of global malaria, they cannot be used to give reliable predictions about the future. Instead, an alternative two-step statistical approach to mapping vector-borne diseases gave a better description of the present global distribution of *falciparum* malaria and predicted remarkably few future changes, even under the most ex-

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PROTEOME SYSTEMS AND SHIMADZU BIOTECH COMPLETE 1ST STAGE OF THE CHEMICAL PRINTER DEVELOPMENT

October, 2001, Proteome Systems and Shimadzu Biotech have today announced the successful completion of the initial stage of their joint Chemical Printer project. Following this early conclusion to its research and development period, the project will be fast-tracked to full commercialisation. This progress represents a breakthrough in high-throughput proteomics.

Chemical Printing refers to a revolutionary technology for protein identification and characterisation. The basis of the technology is microfluidic printing onto proteins, which are then arrayed onto membranes. In the case of the Chemical Printer, preliminary steps involve the arraying of proteins using Proteome Systems' new gel chip technologies. Employing current methods, the entire sample in a protein array needed to be sacrificed in order to conduct its analysis and identification. The Chemical Printer, which exploits state-of-the-art nanotechnology fluidics, enables microprinting on just a fraction of a protein spot. This allows further study on the remainder of the sample, which could prove invaluable for precious clinical samples.

The collaborators expect the Chemical Printer to become one of the pivotal technologies evolving from The Proteomics Alliance (Shimadzu Biotech, Proteome Systems and Sigma-Aldrich). Proteome Systems plan to integrate the Chemical Printer into its' ProteomIQTM platform for high-throughput proteomics. Furthermore, the platform will also be sold as part of the Shimadzu Biotech product portfolio as a stand-alone product and in combination with the Shimadzu Biotech range of mass spectrometers.

Keith Williams, CEO of Proteome Systems, said, "Our team, led by Andrew Gooley, has taken Proteome Systems' patented technology and shown that it performs in a way that is more efficient and flexible than existing in gel digestion technologies. We believe that the Chemical Printer will become a defining instrument in any large scale proteomics program."

Tetsuo Ichikawa, Chairman of Shimadzu Biotech, commented, "The collaboration between Proteome Systems and Shimadzu has put the Chemical Printer project ahead of schedule. This technology is a key to archiving and analysing individual human proteomes, which we believe will be an essential tool in the future."

"The progress of the Chemical Printer project is extremely encouraging and will enable Shimadzu Biotech to provide new solutions in Proteomics when packaged with the Axima MALDI mass spectrometers" remarked Chris Sutton, Business Manager for Shimadzu Biotech's series of MALDI MS products.

Notes to Editors:

About Proteome Systems:
Proteome Systems, with 75 employees in Sydney and Boston, is one of a small group of global companies with the capacity to conduct large-scale proteomics. Proteome Systems uses its own instruments, consumables and informatics technologies as well as partnering with leading technology companies. Proteome Systems is a leading innovator in the development of a high-throughput proteomics platform and, along with its technology partners, anticipates the release of the platform in the second half of 2001.

About Shimadzu Biotech
Shimadzu Biotech, is a new business unit of Shimadzu Corporation. It has been created to unite a

strong solutions-based offering and accelerate the progress of biotechnology research and development. Shimadzu Biotech captures, within one dedicated organisation, the best expertise and technology from within Shimadzu Corporation and its subsidiary, Kratos Analytical.

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